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(71) Applicant (for all designated States except US): MOLECULAR DYNAMICS, INC. [US/US]; 928 East Arques Avenue, Sunnyvale, CA 94086 (US).

(72) Inventor; and

(75) Inventor/Applicant (for US only): MAHTANI, Melanie, M. [CA/US]; 192 Everett Avenue, Palo Alto, CA 94301 (US).

(74) Agent: RONNING, Royal, N., Jr.; Amersham Pharma-BioTech, Inc., 800 Centennial Avenue, Piscataway, NJ 08855 (US).

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(54) Title: ROLLING CIRCLE AMPLIFICATION ASSAY FOR NUCLEIC ACID ANALYSIS

(57) Abstract: Method and reagents for analysis of nucleic acid sequences is disclosed. In this method a plurality of padlock probes is provided. Each padlock probe may hybridize to a locus on a target nucleic acid under hybridization conditions. If a targeted variant is present at the locus, the padlock probe may be ligated to form an amplification target circle. The amplification target circle acts as a template for production of tandem-sequence DNA. The tandem-sequence DNA may then be digested into non-tandem detection fragments which are subsequently separated and detected. The plurality of padlock probes are designed such that ligation of the probes, amplification of the target circle, and digestion of the tandem-sequence DNA subsequently produced, and detection may all be effected with the same set of reagents. Each probe targets a unique locus variant on the target nucleic acid sequence and produces a detection fragment that may be distinguished from detection fragments produced from other padlock probe in the plurality of padlock probes by using a fragment analysis detector.

## INTERNATIONAL SEARCH REPORT

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**A. CLASSIFICATION OF SUBJECT MATTER**  
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According to International Patent Classification (IPC) or to both national classification and IPC

**B. FIELDS SEARCHED**

Minimum documentation searched (classification system followed by classification symbols)

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Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)

EPO-Internal, MEDLINE, BIOSIS, WPI Data, PAJ

**C. DOCUMENTS CONSIDERED TO BE RELEVANT**

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	LIZARDI P M ET AL: "MUTATION DETECTION AND SINGLE-MOLECULE COUNTING USING ISOTHERMAL ROLLING-CIRCLE AMPLIFICATION" NATURE GENETICS, NEW YORK, NY, US, vol. 19, no. 3, July 1998 (1998-07), pages 225-232, XP000856939 ISSN: 1061-4036 the whole document	1-11
A	WO 00 04193 A (UNIV YALE) 27 January 2000 (2000-01-27) page 15 page 24-25 page 53-58; claims	1-28
A	US 5 854 033 A (LIZARDI PAUL M) 29 December 1998 (1998-12-29) column 1-3; claims; example 5	1-28
-/-		

Further documents are listed in the continuation of box C.

Patent family members are listed in annex.

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Name and mailing address of the ISA

European Patent Office, P.B. 5618 Patentlaan 2  
NL - 2280 HV Rijswijk  
Tel. (+31-70) 340-2040, Tx. 31 651 epo nl,  
Fax. (+31-70) 340-3016

Authorized officer

Reuter, U

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## C(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	WO 99 09216 A (UNIV ROCHESTER) 25 February 1999 (1999-02-25) page 6-9; figures 1,2 page 34-35 -----	1-28

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